

# GENETIC STUDIES OF TWO SPECIES OF INDIAN CATFISHES

ABSTRACT

ALKA (TANDON) PRAKASH

DEPARTMENT OF ZOOLOGY

SCHOOL OF LIFE SCIENCES

REGISTRATION NO. 406

THESIS SUBMITTED IN FULFILMENT OF THE REQUIREMENT OF  
THE DEGREE OF  
DOCTOR OF PHILOSOPHY

TO



NORTH-EASTERN HILL UNIVERSITY

SHILLONG, INDIA

APRIL, 1991

Air-breathing fishes can use atmospheric oxygen directly for respiration, besides their gill-respiration. They are marketed alive and are also referred to as "live fishes". Both Clarias batrachus and Heteropneustes fossils are readily available in the North-eastern part of India and they fetch a high price due to their nutritive value.

The present genetic work on both these catfishes is biochemically oriented. It has been divided into various sections, including introduction, materials and methods, observations, discussion and concluding remarks. In each of these sections we have included the study of four dehydrogenases, that is, lactate dehydrogenase, malate dehydrogenase, alcohol dehydrogenase, and glucose-6-phosphate dehydrogenase, in at least six tissues from these fishes. Related work has been referred to in the introductory section.

Live fishes were sacrificed in the laboratory and fresh tissue samples from the kidney, liver, eye, muscle, brain, heart and gonads were used. All the experiments were carried out in a cold condition, below

4 degrees centigrade. In order to study the isozymes of the above mentioned enzymes the technique used was polyacrylamide gel electrophoresis followed by specific staining procedures. Distinct bands were obtained on the rod gels which were photographed and are presented in the various plates. Heat inactivation studies were also carried out. The bands were characterised and related to the genetics of these fishes.

Lactate dehydrogenase isozymes can be visualised on the gel due to the difference in their net charge. In most vertebrates this tetrameric enzyme is coded by two gene loci, LDH A and LDH B which synthesise two subunits A and B. The random association of these subunits yields five isozymes. The A subunit predominates in tissues under anaerobic glycolysis like the skeletal muscle and the B subunit in tissues with aerobic metabolism like the heart. Teleostean fishes possess a third locus the LDH C locus in addition to the LDH A and LDH B loci. The C subunit containing isozymes are synthesised by the neural tissues or the digestive tissues, predominantly in the liver. In Clarius batrachus and Heteropneustes fossilis at least three

LDH loci, the LDH A,B, and C loci, code for the observed isozyme patterns. Tissue-specific expression of the various isozymes is observed and a reverse mobility of the A and B isozymes is seen, as compared to most other vertebrates. In C-batrachus the cathodal-most kidney specific band, K, is seen to be present in the brain and heart tissues as well. This is distinct from the liver specific, C/F, band in its mobility. The K subunit differs distinctly from the C subunit, the former having a much higher molecular weight than the latter, as is observed from the detailed experiments in our present work. The expression of both the kidney-specific and the liver-specific bands in an individual gives us a clue to the idea that there might be more than three loci at play in this fish resulting in the unique pattern of the LDH isozymes observed therein.

Malate dehydrogenase is a dimeric enzyme catalysing the inter-conversion of malate to oxaloacetate in the Krebs cycle. It exists in two forms, the supernatant MDH(s-MDH) and the mitochondrial MDH(m-MDH). We have used the s-MDH which is known to be dimeric in nature and is coded by two gene loci, the MDH

A and the MDH B loci. Most tissues in our fishes exhibit a three band pattern along with the expression of some allelic forms. The kidney and liver tissues are sites of high MDH activity. There is evidence for the activity of an allele, the A', in these fish along with the A and B loci of MDH.

Alcohol dehydrogenase is primarily a liver-specific protein. It catalyses the interconversion of many alcohols to their corresponding aldehydes and ketones in the presence of NAD(H). In most fishes it is encoded by a single locus, its distribution varying from tissue to tissue. An overall analysis of this dehydrogenase in both Clarias batrachus and Heteropneustes fossilis shows the existence of at least two loci coding for the observed isozyme patterns. In C-batrachus three bands are scored in the eye tissue and in H-fossilis the maximum number of band phenotypes scored in the various tissues has been six. We have postulated two different models showing the phenotypic expression of various loci in our fishes. In C-batrachus two gene loci, A and B, code for the observed isozyme patterns whereas in H-fossilis an additional

allele, the A' gene, along with the A and B loci is responsible for the ADH activity observed in the various tissues of this fish.

Glucose-6-phosphate dehydrogenase catalyses the glucose-6-phosphate oxidation, providing NADPH for biosynthetic purpose, in the pentose phosphate pathway. G6PD isozymes in mammals are encoded in two separate loci, one locus being sex-chromosomally linked and the other being autosomal. The latter reacts more actively with galactose-6-phosphate and is now referred to as Hexose-6-phosphate dehydrogenase. Two forms of G6PD, called A and B, are also found in Clarias batrachus. The A fraction corresponds to G6PD and is observed in all the tissues, exhibiting high activity of this enzyme. The form corresponding to H6PD is observed only in the liver tissue. Here, G6PD is encoded by a single locus resulting in a single band as is observed in the various tissues of this fish. In Heteropneustes fossilis the brain and eye tissues exhibited a five band phenotype, whereas in other tissues G6PD activity was observed as a single band. In this fish G6PD may be a tetrameric enzyme resulting from the products of two codominant

autosomal gene loci. No sex differential G6PD activity was observed in both these fishes.

In the concluding remarks we have suggested what work can be done in the future, on the same lines, in order to confirm and fill the lacunae in the present work. This would involve a population genetic study related to the ecology of these fish.

We have also made a morphometric and meristic study on these fish. This has been included in Appendix I. This was done mainly because during the course of our research we came across some unique individuals of C. batrachus which had a distinct spotted appearance and a pinkish cream body colouration. Various morphometric parameters have been taken into account and regression equations of these parameters related to the entire body length have been presented in various tables. Length-weight relationships and the relative length of the gut of these fishes have also been analysed.

In Appendix II we have dealt with the karyotypic study of the chromosome complements of C.

batrachus and H. fossilis. There has been some controversy regarding their  $2n$  number which has been confirmed in the present study. We have found that in C. batrachus,  $2n=52$  and in H. fossilis,  $2n=56$ .

**LEHU LIBRARY**

Acc. No

Acc by

Date

Class by

Sub Heading

Enter by

Inscribe